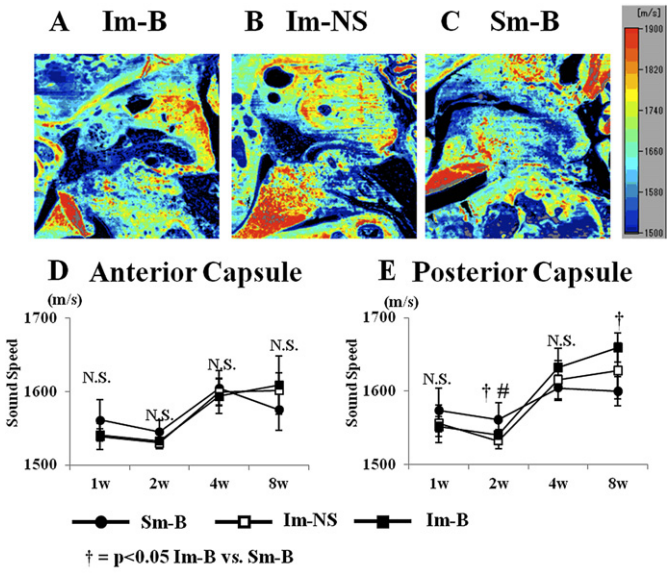

Figure 3. A- C: Posterior Capsule at 8 week.
Low sound speed area: Black to blue, High sound speed areas: Yellow to red.
D and E: The sound speed of the capsule.

127
THE EFFECTS OF DELAYED ADMINISTRATION OF RHO-KINASE INHIBITOR FASUDIL ON SURGICALLY INDUCED OSTEOARTHRITIS IN RATS

M. Pest, V. Pitelka, F. Beier. *Univ. of Western Ontario, London, ON, CANADA*

Introduction: Osteoarthritis (OA) is a highly prevalent joint disease in North America. Current treatment methods focus on relief of symptoms including pain, with little to no effect on the underlying joint damage. Our studies have demonstrated that the signaling molecule transforming growth factor alpha (TGFα) is upregulated in animal models of OA and a subset of human cases. Downstream targets for TGFα include rho-associated protein kinase (ROCK). Inhibition of ROCK in cartilage explant cultures has been shown to decrease catabolic degradation of collagen II and aggrecan. Corroboration of these findings in an animal model would help to solidify ROCK and the TGFα pathway as viable candidates for drug studies in human tissue.
Purpose: To evaluate the protective effects of delayed administration of ROCK inhibitor fasudil (HA-1077) following surgical induction of rat OA in vivo.
Methods: OA was induced surgically in the right knee joint of male Sprague-Dawley rats by method of anterior cruciate ligament transection with partial medial meniscectomy; sham surgery serves as a control. Treatment began at 4 weeks post-surgery using osmotic pumps administering vehicle, 3 or 15 mg/kg/day of fasudil to groups of 5 rats each with an additional sham-operated control group. Groups were terminated at time points 3 and 6 weeks after initiation of treatment, with additional vehicle and sham groups sacrificed at 4 weeks post-surgery as comparisons. Development of OA was evaluated in safranin-O/fast green stained coronal knee sections using a modified OARSI scoring system.
Results: Rats treated with fasudil at a concentration of 3 mg/kg/day for 3 weeks exhibited statistically significant lower histologically assessed cartilage damage compared to vehicle and 15 mg/kg/day treatment groups; this group was also not statistically different from vehicle at 4 weeks post-surgery (0 weeks treatment). This effect was lost at 6 weeks of treatment. Treatment with 15 mg/kg/day fasudil showed no significant difference from vehicle at any time point, or difference from 3 mg/kg/day fasudil at 6 weeks treatment.
Conclusions: Treatment with a low dose of fasudil through subcutaneous osmotic pumps slowed the progression of cartilage damage at 3 weeks treatment time in rats with established OA, however this effect was lost at 6 weeks treatment. A higher dose does not seem to protect against cartilage degeneration at either 3 or 6 weeks treatment, which may be due to toxicity or other dose related effects.

128
IMPROVED ASSESSMENT BY QUANTITATIVE DIGITAL HISTOMORPHOMETRY OF HISTOPATHOLOGICAL CHANGES OF ARTICULAR CARTILAGE IN A SURGICAL MODEL OF POST-TRAUMATIC OSTEOARTHRITIS OF THE KNEE JOINT IN RATS.

K.A. Rudolphi. *Sanofi, Frankfurt-Main, Germany*

Purpose: The goal of the study is to evaluate the development of the histopathological changes over time in a model of surgically-induced osteoarthritis of the knee joint in rats and to compare the most frequently used subjective semi-quantitative scoring of joint damage with computerized quantitative digital histomorphometry.
Methods: Male skeletally mature Lewis rats (12 weeks of age, n = 10-12 animals/group) were subjected to unilateral transection of the Anterior Cruciate Ligament plus 25% removal of the Medial Meniscus (ACLTpMx model) or sham operated. Three, 7, 28, 56 and 84 days following ACLTpMx the animals were sacrificed and in Hematoxylin-Eosin- and Safranin-O-stained (SO) coronal paraffin sections the degree of joint damage was evaluated by two observers in a blinded fashion using a modified histopathological Mankin-score. For quantitative histomorphometry digital images of the joint were analyzed using the digital image analysis software Integrator VIS System Version Nr. 3.0.15.0 (http://www.visiopharm.com Denmark). The degree of cartilage destruction and subchondral bone sclerosis was quantified by measuring the following parameters: 1. cartilage surface irregularity, 2. cartilage area, 3. chondrocyte number, 4. area of proteoglycan-containing (SO-stained) cartilage and 5. area of sclerotic subchondral bone.
Results: The histopathological changes (cartilage fibrillation and erosion, chondrocyte loss, proteoglycan depletion and subchondral bone sclerosis) developed rapidly with increasing severity over time. Already 28 days after ACLTpMx moderate to severe signs of OA were observed. Sham-operated animals did not develop significant OA pathology at any time point. The histomorphometric parameters showed a significant correlation with the corresponding Mankin-subscores.
Conclusions: The ACLTpMx model of OA in rats shows similar features as human knee OA regarding anatomical location and the specific histopathological morphology. Quantitative digital histomorphometry of cartilage destruction and subchondral bone sclerosis offers a more objective and less time consuming assessment of OA histopathology in this experimental model than classical histopathological scoring, thus facilitating the preclinical pharmacological testing of potential disease-modifying drugs.

129
GAIT ANALYSIS AFTER HYALURONIC ACID INJECTION INTO OSTEOARTHRITC KNEE JOINTS OF MOUSE

Y. Muramatsu [1], T. Sasho [1], M. Saito [1], S. Yamaguchi [1], N. Ikegawa [1], R. Akagi [1], S. Mukoyama [1], A. Watanabe [2], Y. Wada [2], K. Takahashi [1]. [1] *Dept. of Orthopaedic Surgery,Graduate Sch. of Med.,Chiba Univ., Chiba, JAPAN;* [2] *Dept. of Orthopaedic Surgery, Teikyo Univ. Chiba Med. Ctr., Ichihara, Japan*

Background: Several experimental animal models of osteoarthritis(OA) have been developed to help our understanding of OA. They are especially useful in histological assessments of interventions against OA but behavioral analysis, which is another important aspect of OA feature, has sparsely been performed on them. Gait disturbance results from joint pain associated with OA and gait analysis would be important to evaluate the progression of OA as well as histological evaluation. In the present study, gait analysis was conducted with CatWalk systemTM developed for the use of small animals. It is an automated gait analysis system and has been validated as a method to quantify abnormal gait pattern in rat models of arthritic pain. But there has been no comprehensive analysis of its use in mouse model along OA development.